VOLUME LXI NUMBER 1

THE BOTANICAL GAZETTE

JANUARY 1916

THE ACTION OF SCHUMANN RAYS ON LIVING ORGANISMS

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(WITH FOUR FIGURES)

The effect of light upon organisms is a subject of steadily growing interest and importance. The work of recent years indicates that we may hope to discover the fundamental principles involved in the action of light on protoplasm more readily by turning attention to the shorter light waves than by continuing to investigate the action of the longer waves which occur in sunlight. The chief reason for this is that light of shorter wave lengths acts more rapidly and produces chemical and structural changes in such a way that its action can be much more readily followed. While these changes may not be identical with those produced by light of longer wave lengths, it may be taken for granted that the knowledge gained by studying the effects of the shorter light waves will prove to be of the greatest assistance in explaining the action of the longer waves.

The present paper is a report of some observations by the writer on the effects upon protoplasm of light in the Schumann region of the spectrum, the region containing wave lengths between 2000 and 1250 Ångström units. This region of the spectrum is of particular interest because the light which it contains has a much more injurious action upon protoplasm than has the light of longer wave length.

No previous investigations have been made of the effects upon protoplasm of light in this region of the spectrum.[1] One reason for this is that the employment of Schumann rays presents many difficulties. A special technique is required, and sources of these rays suitable for biological investigations have been lacking. The investigations on Schumann rays described in this paper were made possible by the kindness of Professor THEODORE LYMAN, who placed at the disposal of the writer the necessary apparatus and pointed out the methods by which the difficulties of technique might be surmounted.

The effects of ultra-violet light upon a variety of organisms have been studied and an attempt has been made to follow as carefully as possible the changes produced in the protoplasm. The changes in structure have been followed by observing the organisms under the microscope during the action of the ultra-violet light. The comparative efficiency of waves of various lengths in producing these changes has been studied by a variety of methods.

As previously stated, it has been found that Schumann light is much more injurious to protoplasm than any light heretofore reported. An exposure of only a few seconds to the light from a comparatively feeble source is sufficient, not only to bring about death, but also to cause a complete disorganization of the protoplasm. It has further been found that the destructive action is not lessened when the organism is exposed while it is thoroughly desiccated and in a high vacuum. As will be pointed out later, this fact is significant, for it indicates the direction in which we must look for an explanation of the changes produced by these and other electromagnetic waves.

While no record of previous investigations of the biological effects of Schumann rays has been found, it seems advisable to give a brief review of certain investigations relating to the biological effects of light of longer wave lengths.

Taking sun baths for hygienic purposes is a very ancient and widespread practice. The *heliosis* and the *insolatio* were

[1] BILLON-DAGUERRE (Compt. Rend. **149** and **150**: 1909) claims to have sterilized liquids by the Schumann rays. He made no study of the biological effects of the rays, and, moreover, it is by no means certain that his results were due to the action of Schumann rays. On this point see LYMAN (**28**).

important features of the public baths of the Greeks and the Romans.

In the beginning of the nineteenth century VALLET (33) reported a complete cure of a case of dropsy by exposing the patient to sunlight one hour each day for 14 days, and LÖBEL (24) cured a case of amaurosis by focusing sunlight upon the diseased eye. LÖBEL thought that the beneficial effects of the exposure were due, not only to the physical action of the heat and the light, but also to the chemical action of the sunlight.

In 1858 CHARCOT (9) reported the experiences of two chemists who were using an electric arc for vitrifying certain materials. The electric current was obtained from a Bunsen pile of 20 elements. The experiment lasted one and one-half hours. The experimenters were 50 cm. from the arc, and experienced no change in temperature, but their eyes pained them so severely the following evening and night that they were unable to sleep. The next day they had a painful erythema of the skin. CHARCOT cites other cases of electric light burn caused by an arc from a battery of 600 Bunsen elements. He concluded that the erythema was due to the chemical action of the light, and not due to the heat.

The first impetus to a critical study of the destructive action of light came when the germ theory of disease had been thoroughly established and scientists had recognized the importance of discovering efficient methods of disinfection. In 1877 DOWNES and BLUNT (10) reported the results of their experiments on the effect of light upon bacteria and other organisms. They undertook an investigation to determine whether or not light has a deleterious effect upon bacteria. They exposed culture media contained in glass tubes to sunlight. They showed that light is inimical to and, under certain conditions, may wholly prevent the development of organisms which are prone to appear in culture media. The action is more energetic upon bacteria than upon mycelial fungi. The fitness of the substratum for growing bacteria is not impaired by insolation. By using colored screens they showed that the blue end of the spectrum is more active than the red end. In a second paper (11) they showed that light will kill organisms which are in distilled water, and organisms which are air-dried. They tried to

approach the problem of the mechanism of the light action by studying the action of light upon organic substances. The study of the action of light upon oxalic acid showed that by insolation oxalic acid is decomposed into carbon dioxide, carbon monoxide, and water. They found that oxygen is necessary for this decomposition. Experiments were then conducted on the action of light upon ferments. They showed that light, in the presence of oxygen, destroys the power of yeast to ferment sugar. They had intended to make an exhaustive study of the action of light upon organic compounds, but CHASTAING preceded them, showing that many organic substances were oxidized by the light when in the presence of free oxygen.

DUCLAUX (**15**) in 1885 studied the action of light upon the anthrax bacillus. He was the first to study the action of light upon pure cultures of bacteria. He found that the ability of the organisms to resist the action of the light varies with the species, with individuals within the species, and with the nature of the culture medium.

In 1887 ROUX (**30**) found that his culture media, when exposed to sunlight, became toxic to the spores of the anthrax bacillus. DUCLAUX had previously shown that carbohydrates are easily oxidized in sunlight. ROUX concluded, therefore, that his culture media were rendered antiseptic by the oxidation of the carbohydrates which they contained.

WARD (**34**) in 1892 separated the action of the light upon the medium from the action of the light upon the organism by exposing the spores in a thin film on glass and then adding agar. The exposed spores were killed. In other experiments he first exposed the agar to the light, and then added it to thin films of unexposed spores on glass. The unexposed spores grew in the exposed agar. He attributed the action of the light to the destruction, in the presence of oxygen, of fatty foods stored in the spores. WARD in later papers (**35, 36, 37**) described experiments on the relative toxicity of the various parts of the spectrum. Instead of exposing culture tubes to various parts of the spectrum, as several observers had done before, he exposed a culture evenly charged with organisms (*Bacillus anthrax* and *B. subtilis*) to the spectrum, formed by a

quartz prism, of the light from a carbon arc. He was unable to tell exactly where the effect began, but in general it began at the blue end of the green, reached a maximum in the violet end of the blue, and diminished again in the violet and ultra-violet. The action extended into the ultra-violet. The culture was cooled on ice during the exposure. WARD suggested that light from a naked arc might prove efficient in disinfecting hospital wards, railway carriages, or other places where rays can proceed directly to the organism. He pointed out that a study of the action of light upon cells might teach us much concerning sunburn, sun baths, etc.

Thus far the use of light as an agent for disinfection had not proved practicable, and interest in the destructive effects of light was becoming purely academic, but the subject received a new impetus by the discovery of phototherapy. In 1871 two papers appeared in the *Lancet*, one by BARLOW (**2**), the other by WATERS (**38**), on the deleterious effects of light in the treatment of smallpox. In 1893 FINSEN (**16**) published a paper in which he reviewed the work of CHARCOT, WIDMARK, and HAMMER. A little later he published a second paper on the treatment of smallpox cases in the absence of the chemical rays (**17**). This paper was followed shortly by three others on the same theme, and later in the same year by a paper concerning the destructive action of chemical rays upon animal organisms (**18**). FINSEN'S work, published in 1893, was entitled *Negative phototherapy*.

In 1896 there was held in Copenhagen a meeting of university professors and influential laymen for the purpose of studying the value of light in the treatment of disease. At this meeting FINSEN read a paper entitled "Om Anvendelse of koncentrerede, kemiske Lysstraaler i Medicinen" (Kopenhagen, 1896), which set forth what he called "positive phototherapy," as opposed to his "negative phototherapy" of 1893. As a result of this meeting the "Finsen Medicinske Lysinstitut" was founded. The personnel of the institute consisted of 7 doctors, a physicist, an electrician, and 33 nurses. The results of the research of the institute from 1900 to 1907 were published in the *Mitteilungen* of the institute.

The basis of "positive phototherapy" is given by BIE, a member of the institute, in a paper published simultaneously in medical

journals in England, America, Germany, and France (**3, 4, 5, 6, 7**). The experimental basis of FINSEN's phototherapy is (1) the bactericidal property of the chemical light waves; (2) the power of the chemical rays to produce erythema; (3) the power of the chemical rays to penetrate the skin. The bactericidal property of the chemical rays had been demonstrated by previous workers. WIDMARK had proved by experiment the fact, pointed out by CHARCOT 30 years before, that photoerythema is produced by the chemical rays of light and not by the heat rays. GOODNEFF and FINSEN demonstrated the powers of chemical rays to penetrate the skin, by placing sealed glass tubes containing silver chloride under the skin of cats and of dogs and exposing to light. The silver chloride was blackened. FINSEN also showed that light will penetrate bloodless tissue, but will not penetrate tissue containing blood. He placed strips of sensitive paper on one side of a man's ear and allowed blue and violet rays of concentrated sunlight to fall upon the other side of the ear. After 5 minutes the sensitive paper was not affected, but if the blood was forced out by pressing the ear between glass plates, the paper was blackened in 20 seconds. In agreement with this is the fact that the spectrum obtained by passing light through an ear filled with blood consists of only a red stripe, while the spectrum obtained by passing light through an ear made anemic consists of all colors.

It is worth while to consider in some detail the method used in the practice of FINSEN's phototherapy. A carbon arc, carrying 50–60 amperes, is used as a source of light. Previous to 1901 sunlight which had passed through a concentration apparatus was used for treating the patients. Its use was abandoned because, aside from the uncertainty of weather conditions, it was found that sunlight is not only weak in the extreme ultra-violet region (the therapeutically effective part of the spectrum), but it contains an abundance of light in the blue-violet region. The blue-violet waves so tan the skin that after one or two treatments the deposit of pigment makes further treatment impossible. The carbon arc, on the other hand, emits light of shorter wave lengths than those found in sunlight. These short light waves have a marked action upon the surface layers of the skin. They destroy many of the epidermal

cells, including those which contain the pigment. The skin, therefore, becomes more transparent with each successive exposure, and hence there is a continual increase in the penetration of the light.

Experiments were made with light sources which emit a relatively greater amount of light in the extreme ultra-violet than is emitted by the carbon arc; but it was found that there was no increase in the therapeutic effects, while there was an undesirable increase in the amount of destruction of the epidermal cells (**29**).

The therapeutically effective rays are those which have wave lengths between 4000 and 3220 Ångström units. These rays, after passing through a layer of skin 4 mm. thick, have a strong destructive action upon bacteria. Light of wave lengths shorter than 3220 Ångström units has no action upon bacteria which lie beneath the surface of the skin (**23**).

The light from the carbon arc is passed through a concentration apparatus provided with condensing lenses of quartz, and also with water filters for absorbing the heat rays. The area to be treated is made as nearly bloodless as possible and is exposed to the light for 1 hr. and 15 min. at intervals of 1–3 days. The local anemia is produced by pressing the area during the exposure with a quartz lens. In certain skin diseases, notably *Lupus vulgaris*, the light treatment is so successful that out of 350 cases treated previous to 1899 there were none which did not show improvement, and only 5 which were not cured. The result is so certain and so constant that there is every reason to doubt the accuracy of the diagnosis of *Lupus vulgaris* when the method fails.

Besides developing "positive phototherapy" to a high degree of perfection, the studies carried on at the Finsen Institute contributed a large amount of information to our general knowledge of the destructive action of light. The experiments of previous investigators were carefully repeated and their significance was critically discussed, while extended researches were made into new fields.

As pointed out by LÖBEL long ago, the biological effects of exposure to light are the result of photochemical action. Hence if we are to obtain a clear understanding of the biological action of ultra-violet light, it will be necessary to consider some of the characteristics of photochemical reactions.

Photochemical action necessitates light absorption, although all light absorption is not accompanied by photochemical action. Since, for substances in general, light absorption increases as the wave length decreases, chemical action also increases as the wave length decreases; and, as pointed out in the pioneer work of DOWNES and BLUNT, the destructive action of light upon protoplasm increases as the wave length decreases.

There is evidence for the supposition that the chemical characters of some of the elements are changed when they are acted on by ultra-violet light. For example, when oxygen is acted on by light of short wave length ozone is formed; that is, ozone is more stable in such oxygen than it is in ordinary oxygen. In passing, it may be pointed out that this fact is of particular interest to the biologist, for ozone is more opaque to short light waves than is molecular oxygen, and it seems that life on earth is possible only because the ozone formed in the upper layers of the atmosphere by the ultra-violet of sunlight serves as a light-filter and protects the organisms on the surface of the earth from these shorter and more destructive rays. Chlorine may be mentioned as another example. In this case, not only are the chemical characters of the atom changed, but according to TRAUTZ (32) the specific heat as well.

In consequence of the fundamental nature of the changes produced by the light, it is often found that many compounds containing the same element are photosensitive. For example, light affects many of the compounds containing silver. Protoplasm contains many photosensitive elements, and it is found that protoplasm and a large number of the substances elaborated by protoplasm (sugar, starch, cellulose, chitin, hair, rubber, etc.) are decomposed when exposed to ultra-violet light.

The temperature coefficient of light reactions is very low. For photochemical reactions, therefore, temperature has but little influence upon the speed of the reaction. Photochemical changes may take place in dry materials or in a vacuum. The writer has found that the time required to kill spores of fungi was the same, whether the spores were exposed while in a very high vacuum or while in the air and turgid with imbibed water. This result is most

surprising in view of what we know of biochemical reactions, all of which take place in aqueous media.

DREYER and HANSSEN (**14**) showed that albumins and globulins were coagulated when exposed to ultra-violet light, and the writer (**8**), by an investigation of the temperature coefficient of the reaction, showed that light coagulation, like heat coagulation, involves two reactions: (1) a chemical change in the albumin and (2) the precipitation of the albumin. He showed that the first reaction has a very low and the second a high temperature coefficient.

HENRI (**19**) determined the coefficient of absorption of egg white and found that there is a close parallelism between the absorption by the albumin of the various wave lengths and their destructive action.

A very important phase of the biological effects of light is to be found in connection with the action of the so-called photodynamic substances. The reader is referred to a summary of this subject by TAPPEINER (**31**), as space does not permit a discussion of it here.

The writer has found no published record of previous investigations on the visible effects of the Schumann rays upon protoplasm. Several investigators, however, have made microscopic studies of the visible changes produced in protoplasm by light of longer wave lengths. For the most part such studies have dealt with the effects produced in the tissues of higher organisms, and secondary physiological changes have not been sharply distinguished from the immediate effects of the light. DREYER (**12, 13**) and HERTEL (**21, 22**) have studied the visible effects of ultra-violet light upon unicellular organisms, but neither of these investigators used light which contained the Schumann rays.

In the writer's investigations, described later, the visible effects of light containing the Schumann rays have been studied. The source of light was a hydrogen discharge tube similar to the one described by LYMAN (**25**). The tube had two compartments connected by the internal capillary *D*, fig. 1. This capillary had an internal diameter of about 3 mm. In each compartment there was a ring electrode (*A*) of aluminum. The discharge passing between the electrodes was compressed in the capillary *D*, thus becoming a source of light. The bottom of the tube was closed by the plate *F;*

the top was closed by a transparent fluorite plate *E*. The Schumann rays were emitted through this fluorite plate. As will be seen from the figure, the tube was so designed that the internal capillary was brought as near the fluorite window as possible without excessive heating of the window. This arrangement made it possible to expose the organism very near to the source of light. The discharge tube was excited by a $\frac{1}{2}$-kilowatt transformer manufactured by the Clapp Eastham Company. The transformer worked on a 110-volt alternating circuit of 60 cycles. A variable resistance in the primary circuit was adjusted so that about 0.8 ampere flowed through the primary of the transformer. The current through the discharge tube was 16–18 milliamperes. No capacity, beyond that due to the leads, was included in the discharge tube circuit.

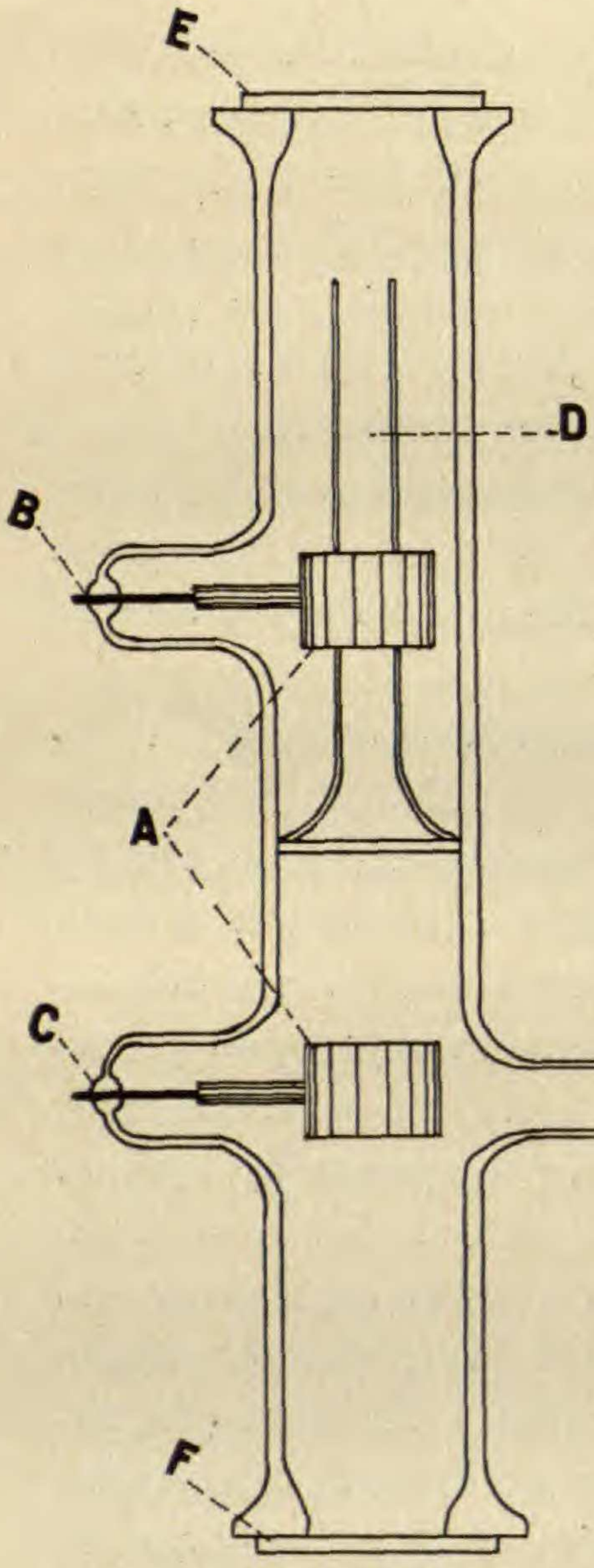

FIG. 1.—Discharge tube used for generating Schumann rays: *A*, ring electrodes of aluminum; *B*, *C*, terminals connecting with a source of high potential (*E*, *M*, *F*); *D*, capillary for increasing the current density; *E*, fluorite window; *F*, glass window.

The primary circuit of the transformer was operated by a relay working on a battery circuit which was controlled by an ordinary telegraph key. The relay circuit had connected with it another circuit which moved the pens on a chronograph. From the chronograph record the exact length of any exposure could be determined. The use of the relay circuit and telegraph key made it possible to operate the discharge tube without the risk of coming in contact with lines carrying currents of higher voltages. This was important when the

tube was used in connection with a compound microscope, for it was often desirable not to look away from the microscope while operating the discharge tube.

The discharge tube was placed upright under the stage of a compound microscope, in the place usually occupied by the condenser and other substage attachments, with the fluorite window flush with the upper surface of the microscope stage. When the discharge tube was in this position the microscope mirror could not be used. Hence it was necessary to illuminate the objects under observation by some other means. Various methods were employed: an arc lamp was placed beneath the work table, and by means of mirrors and lenses a beam of parallel light was directed up through the discharge tube; or the objects were lighted from above, either by concentrating the light on the microscope stage with a condensing lens or by using a special vertical illuminating objective.

The discharge tube was held by a mechanical support so arranged that by moving a lever the discharge tube moved down and away from the microscope stage. The regular substage attachments could then be swung back into operating position. The change from the discharge tube to the substage attachments or from the substage attachments to the discharge tube could be made very quickly, and without interrupting observations through the microscope.

The Schumann region of the spectrum is a region of general absorption for most substances. But few solids are known which transmit even the longest Schumann waves (**26**). Air absorbs all except the longer waves, the absorption being due to the oxygen (**27**). Fluorite is the only substance known which transmits the entire Schumann spectrum. In fact, the Schumann spectrum extends in the direction of short wave lengths only as far as fluorite transmits. It is evident, therefore, that if we wish to expose organisms to the entire Schumann spectrum we can have no substance other than fluorite between the organism and the source of light. Even air must be displaced by the more transparent fluorite. Occasionally the organisms were placed directly on the fluorite window of the discharge tube; more often a special slide was used. The special

slide was a regular microscope slide with a hole 1.5 cm. in diameter bored through it. A disk of fluorite was cemented into the hole with its upper surface flush with the upper surface of the slide. This slide was held in the regular mechanical stage of the microscope and the fluorite window of the discharge tube was thus brought into contact with the fluorite disk in the slide.

When the tube was excited for any great length of time it became hot, and sufficient heat was conducted to the microscope slide to vitiate the results. It was found, however, that the light was so destructive that during a single exposure of sufficient length to kill the organisms, the temperature did not increase more than 1° C. The discharge tube was moved away from the microscope slide immediately after each exposure. The temperature of the drop of water which contained the organisms was measured by means of a thermal junction made of copper and constantin. The sensitiveness of the galvanometer used was such that, with these junctions, one division on its scale corresponded to 0°.05 C. The constant junction was kept packed in ice in a thermos bottle. The variable junction was flattened out very thin and was attached to a flexible support in such a manner that it could be placed beside the organisms under the cover glass. The junction was held in place on the slide by the capillary pressure of the cover slip, and was in the field of view of the microscope during the entire experiment. If the temperature of the drop of water was raised more than 1° C. by the exposure to the light, the experiment was discarded. The arrangement of the tube, slide, and thermal junction is shown in fig. 2.

The length of time required for killing varied both with the species and with the individual organisms. In general, a small organism was killed more quickly than a large one. With a given light intensity, an exposure of several minutes was not sufficient to kill such organisms as rotifers and lumbricoid worms, while *Sphaerella*-like swarm spores, which contain both chlorophyll and an "eye spot," were killed almost instantly. The swarm spores were killed so quickly that there was not sufficient change in temperature to be indicated by the thermal junction. In some of the experiments the intensity of the light was reduced until an exposure

of 10 seconds was required to kill the swarm spores. Exposures of one second duration were then made at intervals of several seconds. It was found that the action of the Schumann rays is additive. The swarm spores were killed only when the total exposure equaled 10 seconds. Other organisms gave similar results. The fact that the action of the light is additive made it possible to interrupt the exposure from time to time, and to make a detailed study of the progress of the changes produced by the light. The protoplasm of the swarm spores which had been killed by the light had a granular appearance. Often some of the protoplasm was extruded from the cells and was rounded up into drops.

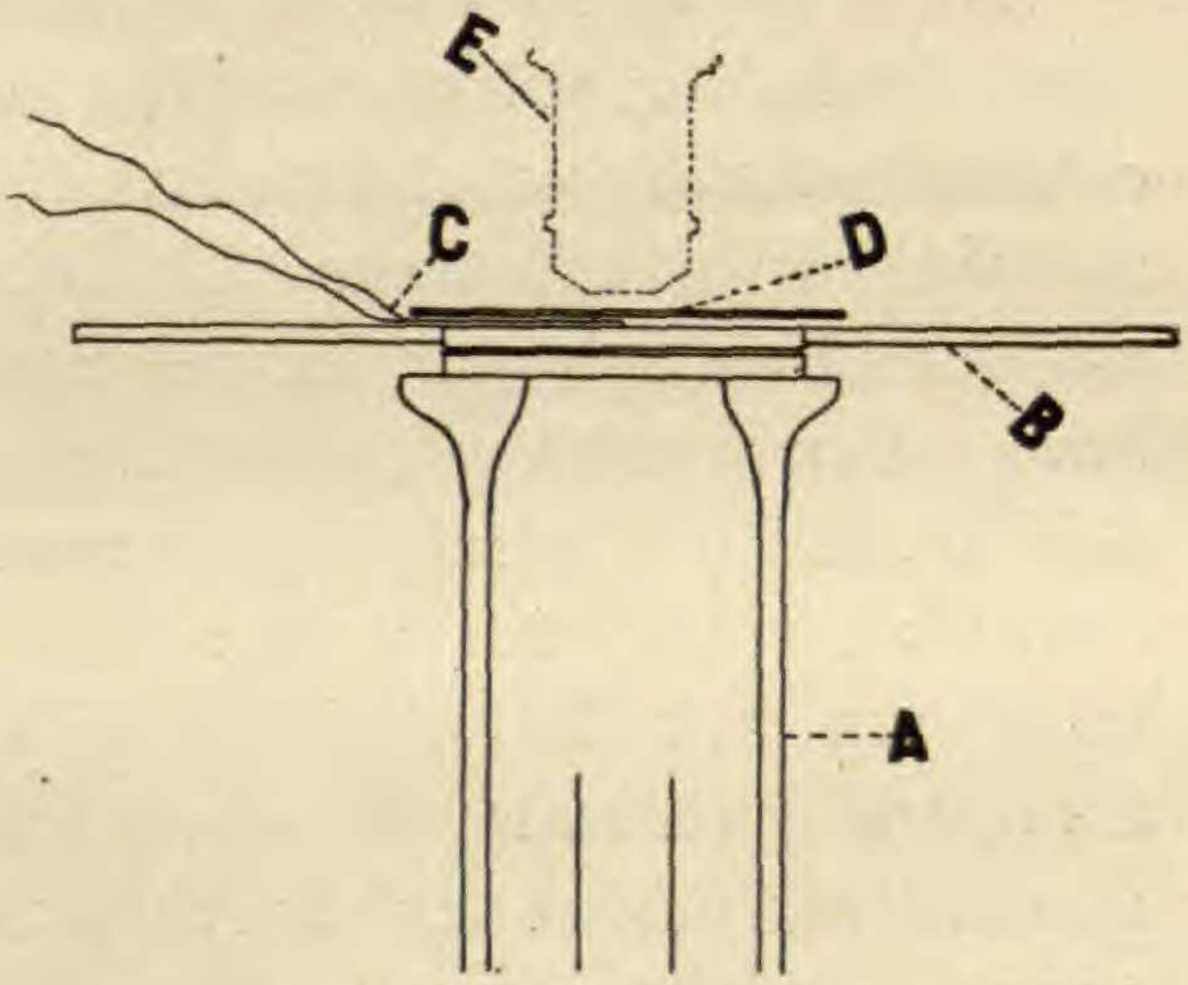

FIG. 2.—Arrangement of apparatus for microscopic observation of the effects of Schumann rays: *A*, discharge tube; *B*, microscope slide containing a fluorite window; *C* thermal junction; *D*, cover glass; *E*, microscope objective.

The cells of a large *Spirogyra* of the *crassa* type were killed by an exposure of 45 seconds when the discharge tube was carrying 18 milliamperes. The first visible change was the disappearance of the wavy margin of the chlorophyll bands. This began on the side of the cell nearest the light. Later the bands broke into isolated rounded drops, each drop containing a pyrenoid. At the same time that the bands were breaking up, they became shorter and contracted around the nucleus. As they contracted, they moved away from the cell wall, pulling the protoplasm lying next to the wall out into threads of viscous appearance. The nucleus became swollen and distended.

When an active amoeba was exposed to the light of the hydrogen discharge tube there was a momentary cessation of motion, followed by a withdrawal of the advancing pseudopodia. Locomotion in another direction began again at once, before the pseudopodia were entirely withdrawn. The extended pseudopodia often turned directly upward away from the light of the discharge tube, and

usually a pseudopodium was sent upward from the upper surface of the body. This pseudopodium was often seen to flatten out against the cover slip. The nucleus moved up into the upper part of this pseudopodium. In some cases so much of the protoplasm flowed up into the pseudopodium that the amoeba became top-heavy and toppled over. One amoeba was seen to send up a pseudopodium, to fall over, and then to repeat the process three times before it was killed. These reactions are really negative phototropic responses, the amoeba moving upward away from the light.

As the protoplasm flowed up into the vertical pseudopodium a thick hyaline ectoplasm was left below. The ectoplasm usually constituted the greater part of the lower half of the amoeba. Often the amount of ectoplasm increased until it nearly equaled the amount of endoplasm. In one case the cover glass was pressed down on an amoeba while in this condition, and the endoplasm and the ectoplasm separated and rounded up into separate drops. Under a magnification of 2200 diameters the ectoplasm showed a few small granules in Brownian motion, but showed no vacuoles. After a prolonged exposure there was often a peculiar flowing of the granular endoplasm out into the ectoplasm. It did not appear to be the same kind of motion that one observes in the regular streaming of the protoplasm, but it was not easy to say wherein the difference lay. After this all motion ceased and the protoplasm appeared coagulated. Under a high magnification (2200 diameters) the protoplasm was seen to be filled with fine vacuoles which were so numerous that it was converted into a fine froth. These vacuoles were not visible before the organism was exposed to the light.

It often happened that only a part of an amoeba was killed; for example, in one case an amoeba which happened to be near a bit of opaque substance when the exposure was made sent a pseudopodium up on top of the opaque substance. The nucleus was next sent up, and then as much of the granular protoplasm as possible. The bit was not large enough to protect the whole organism, and a fringe of protoplasm (ectoplasm) extended beyond it all the way around. The exposure was continued until this fringe was killed. After the exposure, the unexposed part of the organism moved away, leaving the dead fringe behind. In another case the light

was allowed to act for a few seconds on an amoeba which was moving very rapidly across the field of the microscope. The amoeba became quiet during the exposure. As soon as the light was turned off, motion was resumed, but only part of the amoeba moved away; a part of the protoplasm was coagulated and was left behind. The exposure was continued in this way, a few seconds at a time, killing a part of the amoeba at each exposure, until only the nucleus and a small mass of surrounding protoplasm remained alive. A final exposure killed this. The length of exposure necessary to bring about these changes varied from 30 to 100 seconds with the hydrogen discharge tube carrying 29 milliamperes. As previously stated, the entire exposure was not made at one time, but at intervals, so that the experiment often extended over an hour. The changes produced by the light could thus be more carefully observed.

Infusoria are very quickly cytolyzed by the rapid vibrations of these ultra-violet rays. The nature of the cytolysis varies greatly with the species, and in some of the minor details it varies with the different individuals. The writer has observed three kinds of photocytolysis in ciliated infusoria: (1) a cytolysis which is accompanied by the formation of vesicles on the surface; (2) a cytolysis in which some of the internal portions of the protoplasm coagulate; and (3) a cytolysis in which some protoplasm disintegrates directly. The first two types of cytolysis were observed in *Colpoda*-like forms, and the third type was observed in *Stylonychia*.

1. *Cytolysis by the formation of vesicles*.—The cytolysis is, in general, like that of *Paramoecium* in distilled water, in weak alkali, and in 5 per cent alcohol, as described by WULZEN (39). When a *Colpoda*-like infusorian is exposed to the light from the discharge tube carrying 18 milliamperes there is first an increase, then a decrease, in the rate of motion of the organism. Soon vesicles filled with a clear liquid begin to form on the surface of the animal. The infusorian loses its original shape and swims in circles. A vesicle may continue to grow until it is larger than the original organism, or it may increase in size for a short time and then slowly shrink and disappear. As one vesicle is shrinking, others may be forming at some other part of the surface of the organism. If the exposure is

not continued too long the vesicles may entirely disappear and the organism apparently recover. A longer exposure causes the inner wall which separates the organism from the vesicle to rupture and the protoplasm to flow out into the vesicle; while a still longer exposure may cause the outer wall of the vesicle to rupture, permitting the protoplasm to flow out into the surrounding water, with which it is miscible. Sometimes the protoplasm disorganizes and rounds up into drops before it flows into the vesicle. This type of photolysis requires a total exposure of about 30 seconds.

2. *Cytolysis in which parts of the protoplasm coagulate.*—In this type of cytolysis an exposure of 10 seconds causes small areas of the protoplasm to coagulate. The coagulated masses move to the side of the organism and are extruded at once. A longer exposure causes more masses of coagulum to form. As the exposure continues, the masses of coagulum form faster than they are extruded. A swelling appears on one side of the body, which increases in size and then bursts, allowing the protoplasm to flow out into the surrounding water.

3. *Cytolysis in which the protoplasm disintegrates directly.*—When *Stylonychia* is exposed to the light from a hydrogen discharge tube excited by a current of 18 milliamperes, the organism is stimulated and its rate of motion is increased. It then loses its power of coordination, moves about in circles for a time, and finally comes to rest with its cilia still vibrating. Suddenly the outer membrane breaks at some point and a little protoplasm squirts out. Then, starting from this point, a wave of disintegration passes over the organism, leaving the protoplasm in isolated rounded drops. The drops show surface tension against each other, and also against the fluid in which they lie; but a further exposure may cause some of them to unite. If the discharge tube is excited by a stronger current, 50–70 milliamperes, the cytolysis begins at once before the loss of coordination occurs. Cytolysis begins at the posterior end of the organism. The infusorian darts across the field, leaving behind it a trail of its cytolyzed protoplasm. It continues its motion until only a very small amount of the original protoplasm remains intact, and this cytolyzes at the instant motion ceases.

When the dry spores of *Monilia* sp. are exposed to the light no visible change is observed. If, however, after exposure the spores are allowed to absorb water they become turgid, but their protoplasm assumes a coarsely granular, coagulated appearance, which is quite different from the finely punctate appearance of turgid unexposed spores. When turgid spores of *Monilia* are exposed to the light two kinds of changes are observed: either the protoplasm takes on a coagulated appearance, after which no further change is seen, or the spore wall suddenly bursts and some of the protoplasm squirts out with such force that the spore is driven backward by the reaction. The protoplasm, both outside and inside the spore wall, appears granular. Approximately 50 per cent of the turgid spores burst in this manner when exposed to the light. An exposure of 20 seconds, when the discharge tube is carrying 18 milliamperes, is required to cause the spores to burst. A similar squirting out of the protoplasm was observed in a *Navicula*-like diatom, and in the spores of certain water molds when they were exposed to the light.

The fact that the light acts directly upon the organism itself, and not indirectly through the formation of some toxic substance in the medium, was made evident in the experiments in which the drop containing the swarm spores was larger than the window of the discharge tube, so that a few of the spores which were on the outer edge of the drop were not exposed. Those swarm spores which were not exposed to the light were not killed, even though they were at the very edge of the illuminated area. When the exposure was over they often swam into the region where spores had been killed the instant before. As they entered this region they did not make any change either in the rate or in the direction of their motion. From this it may be concluded that the light did not produce any toxic substance in the solution. This conclusion is further strengthened by the fact that occasionally a swarm spore which was within the range of the light from the discharge tube was protected from the direct influence of the light by the shadow of some opaque material contained within the drop. No change which could be attributed to the action of the light was observed in such an individual. Again, the fact that the position of the dead swarm spores

always marked out exactly the outline of the window of the discharge tube may be taken as evidence that no toxic substances were formed in the solution. The observations made on other species of organisms lead to the same conclusion, that the action of the light is on the organism itself.

Notwithstanding the fact that the light was from an exceedingly feeble source, the changes in the organisms were immediate. In the small swarm spores with thin transparent cell walls the changes appeared the instant the discharge tube was excited. It is evident that the Schumann rays are very destructive to protoplasm. The examples just given of the visible effects of these rays upon organisms are sufficient to make it apparent that we are dealing here with a powerful cytolytic agent, and one which warrants further study.

The relation between the wave length and the destructive action of light in the Schumann region of the spectrum has not been previously studied. For the longer light waves this relation has been investigated to some extent, as will appear from the brief summary which follows.

DOWNES and BLUNT, using colored screens, showed that blue light is more destructive to bacteria than red light. WARD, using a quartz prism, confirmed the results of DOWNES and BLUNT, and showed that the killing power extends into the ultra-violet.

Two papers appeared in 1905 on the bactericidal action of light, describing experiments in which not only the wave length but also the intensity of the light was measured. BANG spread the light of a carbon arc into a spectrum by means of quartz lenses and a quartz prism, and measured the relative destructive action of the various parts of the spectrum by determining the length of time it took in a given region of the spectrum to kill an organism (*Bacillus prodigiosus*) growing on the surface of an agar plate. His results showed that, in general, as the wave length of the light decreases the destructive action increases, but that the curve is not uniform, showing a break in the region of wave length 3000 Ångström units. In this region the light is several hundred times less destructive than in the regions on either side, so that the curve shows two maxima. The secondary maximum is in the region of wave length 3500

Ångström units. A measurement of the energy of the spectrum (which was made with a bolometer) showed that the amount of energy decreased with the decreasing wave length; but that in a region nearly coinciding with the region in which the destructive action of the light fell off the amount of energy increased, so that, when the two curves, the energy curve and the destructive curve, are compared, the one is seen to be the inverse of the other. The depression in the destructive curve and the elevation in the energy curve do not quite coincide, but BANG attributed this to a slight shift in some part of his spectrograph.

HERTEL (**21**) used quartz lenses and a quartz prism to form a spectrum of the light from various spark gaps. He measured, by means of a thermopile, the energy of the light of various wave lengths which he allowed to fall upon living tissues. He found that the destructive action of the light varies directly as the energy, and inversely as the wave length. He did not find the two maxima described by BANG.

HENRI (**19, 20**) in 1912 measured the relative destructive action of light of various wave lengths. He used as sources of light a mercury vapor arc in quartz, and spark gaps with cadmium and magnesium terminals. The relative intensity of the light of various wave lengths was measured by the effect upon a photographic plate. He made use of screens for filtering out the various wave lengths. The efficiency of the screens was determined by spectrographic methods. He found that the destructive action of the light increases continuously as the wave length decreases. HENRI did not find a secondary maximum at 3500 Ångström units, as reported by BANG. With the exception of BANG, these investigators have agreed that, in the regions of the spectrum studied, the destructive action of light increases as the wave length decreases. None of their investigations have included the region of the spectrum lying below wave length 2000 Ångström units.

In the experiments described in this paper the relation between the wave length and the destructive action in the Schumann region has been studied. Because of the small amount of energy in the Schumann rays, no attempt has been made to measure the intensity of the light of the various wave lengths. A knowledge of the

relative intensity has been approximated from their effect on a photographic plate. It is known that the spectrum from a hydrogen discharge tube contains a number of bright lines in the neighborhood of wave length 1600 Ångström units, while if the hydrogen is extremely pure there are no lines between wave lengths 2000 and 1675 Ångström units. In this study the destructive action of light including the wave lengths in the region of 1600 Ångström units has been compared with the destructive action of light from which these waves have been filtered out by means of screens. The hydrogen discharge tube previously described was used as a source of light. Three different methods were used.

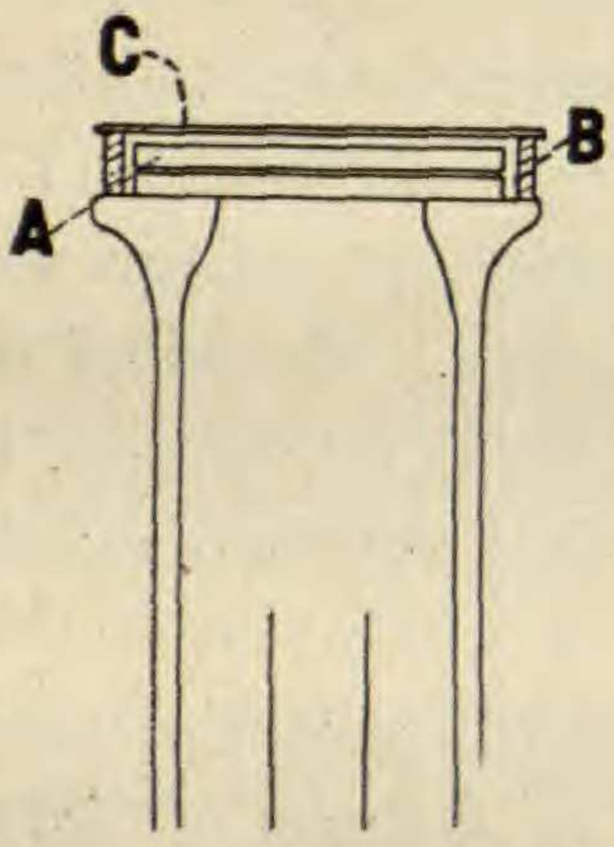

FIG. 3.—Arrangement of discharge tube for studying the relation between wave length and the destructive action of Schumann rays: *A*, rock salt screen; *B*, brass ring; *C*, cover slip.

In the first method the hydrogen discharge tube was placed upright with the fluorite window above. The rock salt screen *A* (fig. 3) was laid upon the fluorite window. Glass plates were coated with nutrient agar and set aside in a sterile closet until the agar became air dry. Spores of *Penicilliun* were placed upon the agar surface, and the plate *C* placed, spores downward, on the ring support *B*. The lower surface of the plate was about 0.05 mm. from the salt screen. After the exposure, the plates were placed in Petri dishes lined with damp filter paper and the Petri dishes set in the incubator. The agar absorbed water and the uninjured spores germinated.

Exposures of various lengths were made, and the shortest exposure which would kill determined. The amount of current flowing through the discharge tube was measured and kept constant during all the experiments. The rock salt screen cut out the light of wave lengths shorter than 1800 Ångström units (**26**). Control experiments were made by removing the rock salt screen and laying in its place a screen of fluorite having the same thickness. The fluorite was transparent to waves longer than 1250 Ångström units. By using the fluorite screen the distance between the spores and the

source of light was kept constant. The short waves would have been absorbed by the air had it not been replaced by the more transparent fluorite.

Spores of *Cephalothecium roseum* were not killed by an exposure of 60 seconds, and spores of a species of *Monilia* were not killed by an exposure of 420 seconds when the screen of rock salt was used, while the spores of both forms were killed by an exposure of 15 seconds when the screen of rock salt was replaced by the screen of fluorite.

In the two other methods the apparatus shown in fig. 4 was used. A glass tube *A*, 31 cm. in diameter, was closed at one end with a glass stopper *B* which was ground in. The other end was closed by a brass ring *C* which had a fluorite disk *D* sealed into its center. The seal was made with De Khotenski cement. A side tube *E* connected the tube *A* with a mercury vacuum pump and a McLeod gauge. The tube was used in a vertical position. An upright brass tube *F* was cemented to the glass stopper. At its upper end it carried a platform *H*. The platform was a copper disk soldered to the head of a screw *I* which passed through a nut soldered to the top of the brass tube *F*. By turning the screw the distance between the platform *H* and the fluorite window *D* could be regulated. A hemisphere *K*, 5 mm. in diameter, was pressed out of a polished platinum plate. Its open side was brazed to a small piece of brass plate *L*.

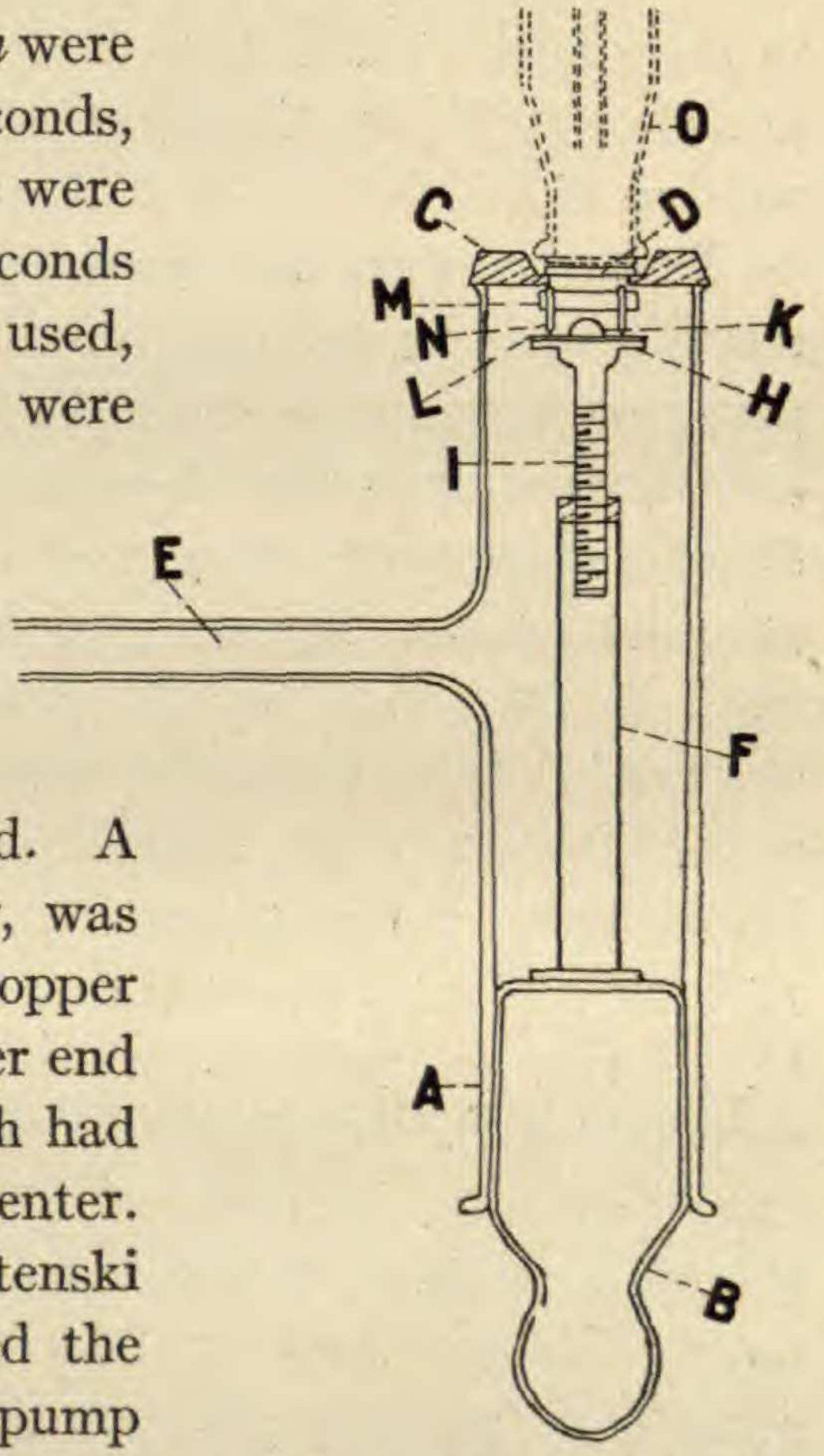

FIG. 4.—Chamber for studying the relation between wave length and the destructive action of the Schumann rays: *A*, glass tube; *B*, stopper; *C*, brass ring supporting fluorite window *D*; *E*, exhaust tube; *F*, adjustable support for the platform *H*; *K*, platinum hemisphere which is brazed to the brass plate *L*; *M*, rock salt screen supported by the brass pins *N*; *O*, discharge tube.

A disk of rock salt *M* with plane parallel faces had holes bored through it into which were inserted brass pins *N*, which served as legs. In operation, these parts were placed in the tube, as shown in the figure. The source of light was a hydrogen discharge tube, as shown at *O*. Its fluorite window was in contact with the fluorite window *D*.

In one method the apparatus was used as follows: The glass stopper with its attached parts was removed from the tube, the platinum hemisphere sterilized in a flame, and then placed convex side upward upon the stage of a binocular dissecting microscope. Fungus spores were transferred to the top of the hemisphere with a platinum needle. By means of the binocular microscope the arrangement of the spores on the platinum hemisphere could be easily observed. After the spores were transferred to the dome a current of air blown through sterile cotton was allowed to play upon the hemisphere. All of the spores which were not in actual contact with the platinum were thus removed. This treatment gave a thin layer of spores quite evenly distributed over a small area at the very top of the hemisphere. The platinum hemisphere was now placed upon the platform *H*, and the screen *M* placed in position, the glass stopper *B* inserted and carefully turned until an air-tight joint was formed. The tube was then exhausted to about 0.001 mm. mercury pressure, and the exposure made. After the exposure, air was admitted into the tube and the stopper *B* removed. The platinum hemisphere with the exposed spores was again placed on the stage of the binocular microscope. A small drop of agar had been allowed to solidify on the under side of the cover slip of a van Tieghem cell. This cover slip was removed from the cell and brought over the spores on the platinum hemisphere. Then, while observation with the binocular microscope was being made, the cover slip was carefully lowered until the "hanging drop" of agar just touched the spores on the top of the hemisphere. The cover slip was immediately lifted, taking the spores with it, and placed back on the ring of the van Tieghem cell. The cell was then set in an incubator. Because of the curved surface of the platinum hemisphere the spores were transferred to the agar without changing their relative positions, and as the agar drop came in contact

only with the spores on the top of the hemisphere, only those which had been directly exposed to the rays of light were transferred. The length of exposure required to kill the spores with and without the salt screen was determined. The results obtained with the spores of *Trichothecium roseum* are given in table I.

TABLE I

Without rock salt screen		With rock salt screen	
Time of exposure in seconds	Percentage of germination	Time of exposure in seconds	Percentage of germination
0	20.5	0	20.0
3	2.9	120	2.0
7	3.2	300	1.0
15	1.3	600	0.0
30	0.0	1200	0.0
60	0.0		
120	0.0		

An unknown amount of light was reflected from the surface of the salt screen. In order to avoid this source of error, the experiments were repeated, and the length of exposure required for killing was determined first with the tube containing air at atmospheric pressure, and then with the tube evacuated. The distance from the top of the platinum hemisphere to the fluorite window *D* was 1 cm. This gave a filtration of 1 cm. of air. LYMAN (**27**) has shown that an air filter 1 cm. thick cuts out all of the shortest Schumann rays. The results obtained with *Trichothecium roseum* are given in table II.

TABLE II

Time of exposure in seconds	Conditions	Results
7	Vacuum	Growth
15	"	"
30	"	Dead
30	Air	Growth
60	"	"
120	"	"
240	"	"
480	"	Dead

I was not able to kill the tan-colored spores of *Penicillium brevicaule* or the black spores of *Stemphylium* sp. (probably *S.*

macrosporoidium). The Schumann rays have not sufficient penetrating power to pass through the colored cell walls.

By this method we were comparing the time required to kill spores in air with the time required to kill them in a vacuum, but preliminary experiments in which the hemisphere *K* was brought very close to the fluorite window *D* showed that the presence or absence of air makes no difference in the length of exposure required for killing. It should be pointed out that for these experiments organisms were selected which had very thin and transparent cell walls. It was impossible to obtain similar results with organisms with thick, dark-colored spore walls. The length of exposure required to kill dark-colored spores was so great that it is doubtful if the Schumann rays took any part in the killing.

The light emitted from the fluorite window of the hydrogen discharge tube is much less destructive when the light waves of a length shorter than 1700 Ångström units are filtered out. The results obtained by the three methods are comparable. The light is 15–20 times more destructive when it contains the short waves than when it does not. The significance of these figures lies in the fact that in the Schumann region, as in the regions of longer wave length, the destructive action of the light increases as the wave length decreases. Necessarily, this statement does not hold true for organisms protected by membranes which are opaque to the Schumann rays.

Summary

By a number of methods it has been shown that the action of the light is on the organism directly, and not indirectly through the formation of some toxic substance in the medium.

It is a well-established fact that the Schumann region of the spectrum is a region in which nearly all substances have strong absorption bands. While no studies have been made upon the absorption of protoplasm in this region of the spectrum, undoubtedly strong absorption does occur. Gelatin, which is a much simpler substance than protoplasm, is so opaque to the rays that the ordinary photographic plate cannot be used in photographing the Schumann spectrum. Special plates, in which the silver salts

are for the most part deposited on the surface of the gelatin, must be used.

HENRI (**19, 20**) has shown that a very thin layer of egg white is opaque to ultra-violet waves between 3000 and 2000 Ångström units in length, and that in this region the opacity increases as the wave length decreases. If the absorption coefficient of egg white can be applied to living protoplasm, and if the opacity of egg white continues to increase with decreasing wave length as we pass from the region studied by HENRI into the Schumann region, then it is reasonable to suppose that the Schumann rays penetrated only a short distance into the substance of the organism. The extreme destructive action of these rays is a result of the strong absorption.

That the rays penetrate only a short distance into the substance of the organism is indicated by the observations made on amoebae, in which only a part of the protoplasm was killed by the exposure to the light. It may have been that the nucleus and the protoplasm which moved up into the vertical pseudopodium were well protected from the shortest waves of the Schumann rays by the thick layer of ectoplasm which remained below. Those parts of the protoplasm which were on the side away from the source of light were killed by the longer, less active light waves only after a prolonged exposure. Again, in the experiments on *Spirogyra*, the visible changes always began on the side of the cell nearest the light. Fungus spores, with brown or tan coloring matter in their cell walls, even though the walls were thin, were not killed by a prolonged exposure to the light. The Schumann rays could not pass through the cell wall.

Because of this strong absorption, the Schumann rays have a marked localized action which gives them a peculiar value for investigations in the experimental morphology and physiology of the cell.

In the experiments on the motile organisms, amoebae and infusoria, it was seen that the Schumann rays have a stimulating effect, to which the amoebae respond by drawing in the pseudopodia and assuming a spherical form, and to which the infusoria respond, first, by an increase in the rate of motion followed by a decrease, then by a loss of the power of coordination, and finally by the disintegration of the living substance.

The examination of highly differentiated cells like those of *Spirogyra* has shown that the visible changes produced by the light are not the same in all protoplasmic structures. The change produced is often one which results in an alteration of the equilibrium of the water content of the protoplasm, as shown by the shrinking and swelling of various parts, by the bursting of spores, and by the miscibility with the surrounding water of the protoplasm of cytolyzed infusoria.

As pointed out in a former paper (8), ultra-violet light causes certain chemical changes in egg albumen, changes which lead to a change in the time-temperature-coagulation curve. A study of the nature of these chemical changes has shown that they result in a decomposition of the albumen molecule. Preliminary experiments upon the effects of ultra-violet light on other protein bodies show a similar destructive action of the light. It would seem, therefore, that the stimulus of light is to be classed with those exciting stimuli which accelerate catabolic changes; and that using, as we have in these experiments, light with high vibration frequencies, we have been able within a short space of time and with no very great light intensity to carry the chemical changes through fatigue and death, and finally to a complete destruction and dissolution of the protoplasm.

The writer has found that spores dried *in vacuo* may be killed by ultra-violet light. This becomes understandable from experiments which the writer made, and which will be published later, which show that albumen and other proteins, dried *in vacuo*, are readily decomposed by ultra-violet light. The effect of these high-frequency electromagnetic vibrations on proteins is comparable to that of dry distillation at high temperature.

These experiments suggest to us a mechanism of the killing action of ultra-violet light, and furnish a clue which, it is hoped, will explain the mechanism of all the effects of light on protoplasm, including those which are not injurious; for it is evident that if the decomposition of the protein molecule is not carried too far it may stimulate the cell without producing injury. A good example of this sort of stimulation is seen in artificial parthenogenesis, which is produced by substances the action of which kills the egg if allowed

to go too far, but merely stimulates it if stopped at the right time.

It is interesting to note that the photolyses previously described follow the photo-chemical-energy law first formulated by TALBOT, that the amount of chemical change is proportional to the product of the intensity times the length of exposure, or, if the intensity is constant, that the amount of chemical change is proportional to the length of exposure. It required the same total length of exposure to bring about cytolysis when the illumination was interrupted as when it was continuous.

In the Schumann region of the spectrum, as in the regions of longer wave length, the destructive action of the light increases as the wave length decreases, and when we consider the very short exposure which was required for killing, notwithstanding the feeble source of light used, it is evident that the light of the Schumann region is much more destructive than the light of the regions of longer wave length. In other words, the curve representing the relation between wave length and destructive action, which slopes upward in the regions of shorter wave lengths, continues into the Schumann region of the spectrum without changing its character.

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